

Kentucky's Experience with Using Diatoms for Bioassessment and Assessing Nutrient Impacts

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Outline

- ◆ History of the Kentucky Algae Program
- ◆ Advantages and Disadvantages of Algae As a Bioassessment Tool
- ◆ Diatom Community vs. Algal Biomass
- ◆ DBI (Diatom Bioassessment Index)
- ◆ Integrating Three Communities in Bioassessment
- ◆ Assessing Nutrients
- ◆ Future Considerations

1977: KY DOW Begins Biological Sampling



It Was Decided to Collect Algal Community Data,



Macroinvertebrate Community Data, and



Fish Community Data



1992: First Used Diatom Bioassessment Index as an Assessment Tool



2000: With New Reference Data Set, DBI Modified to the Present Form



Advantages of Algae As a Bioassessment Tool

- ◆ Algae are attached to the substrate and, therefore, subjected to any immediate or prolonged disturbance.
- ◆ Algae, especially diatoms, are ubiquitous with at least a few individuals found under almost any aquatic condition.
- ◆ Taxonomic richness at any given site is usually high enough for use in calculating various metrics.
- ◆ Diatoms can be identified to the species level by trained phycologists.

Advantages of Algae As a Bioassessment Tool (Cont'd)

- ◆ Autecological requirements (tolerances and sensitivities) are known for many species of diatoms.
- ◆ Benthic algal communities, especially diatoms, have a rapid response and recovery time because of their relatively short life cycle (as compared to fish or macroinvertebrates) and their ability to quickly re-colonize formerly disturbed or impacted sites.

Disadvantages of Using Algae as a Bioassessment Tool

- ◆ Algae samples require a great deal of laboratory processing before they can be analyzed.
- ◆ Algal identification to the lowest taxonomic level requires trained phycologist.

Diatom Community vs. Algal Biomass

- ◆ The diatom community is very sensitive to nutrient enrichment as well as other stressors.
- ◆ Collection of diatom community is easy.
- ◆ Repeatable results can be obtained with duplication of sampling effort.
- ◆ Diatom community data can be utilized with macroinvertebrate and/or fish data to determine bioassessments.
- ◆ Sample processing and analysis are time- and labor-consuming events.
- ◆ Trained phycologist may be difficult to find and hire.

Diatom Community vs. Algal Biomass (Cont'd)

- ◆ Algal biomass can be highly variable within the same reach of a stream. Nutrient enrichment is not the only factor influencing biomass accrual (light, flow, types of substrates, density of mat, etc.). It may be difficult to tease out the effects of nutrients on the biomass from the other factors.
- ◆ It is often difficult to collect and calculate exact coverage area from natural substrates.
- ◆ Sample processing and analysis is less time- and labor-consuming than diatom community structure.
- ◆ Most biologists can be trained to collect, process, and analyze algal biomass data.
- ◆ We just don't have the confidence in the biomass data like we do the diatom community data.

Observational Characteristics of an Algal Community Fully Supporting Designated Uses

Phytobenthos appears diverse with at least four divisions represented, including chrysophytes, chlorophytes, cyanophytes, and rhodophytes. Algal coverage is sparse to moderate (<75%). Phytoplankton sub-community not apparent. Floating algal mats are not extensively present. The algal community is similar to that of reference stations within the same ecoregion or bioregion.

Observational Characteristics of an Algal Community Impacted by Nutrient Enrichment

- ◆ Phytobenthic biomass extensive ($> 75\%$ coverage) dominated by one type of algal growth, such as long filaments of *Cladophora*. Phytoplankton sub-community may be apparent in pools and eddies. Floating algal mats extensive in slow-moving areas. Diversity of algal divisions low with only one or two divisions represented.

Green Filaments (*Cladophora*)



Large Strands of *Cladophora*



Golden Surface Scum



Diatom Bioassessment Index (DBI)

- ◆ Multi-metric index, similar to fish and macroinvertebrate indices, that uses 6 diatom community structure metrics.
- ◆ The DBI is intrinsically designed to be sensitive to nutrient enrichment, as well as other impacts including sedimentation, salinity, acidity, and metals.
- ◆ Box plots were used to measure metric sensitivity between reference and non-reference sites. Pearson's correlation coefficient was used to determine redundancy between metrics.

DBI Development

Original DBI Metrics:

- ◆ Total Number of Diatom Taxa
- ◆ Shannon Diversity
- ◆ Pollution Tolerance Index
- ◆ Percent Sensitive Species

Total Number of Diatom Taxa (TNDT)

An estimate of diatom taxa richness.

TNDT = total number of taxa identified
(those counted and those showing up on the
scan of the slide)

Shannon Diversity (H')

$$H' = - \sum n_i/N \log_{10} n_i/N$$

Where n_i = the number of individuals of taxa i

N = the total number of individuals

Pollution Tolerance Index (PTI)

- ◆ Each taxa is assigned a tolerance value based upon their tolerance to increased pollution.
- ◆ Tolerance values range from 1 (the most tolerant) to 4 (the most sensitive).
- ◆ If not enough information is known about a taxa, a tolerance value of 0 is assigned and that taxa is not counted in the calculation of the PTI metric.

Pollution Tolerance Index (PTI)

(Cont'd)

$$PTI = \sum (n_i \times t_i) / N$$

Where n_i = total # of indiv. of taxa i

t_i = tolerance value of taxa i

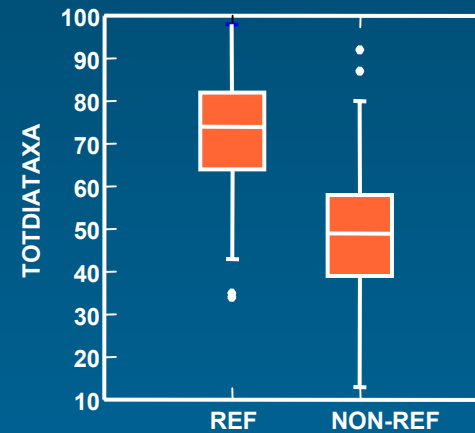
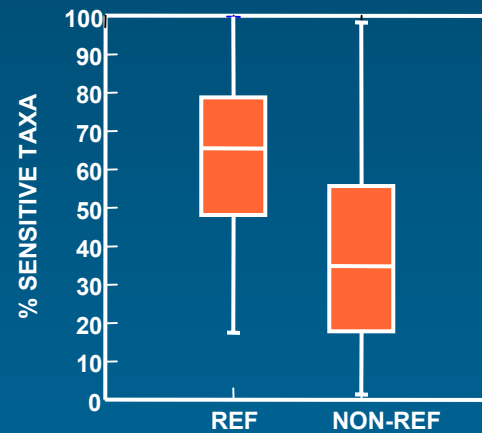
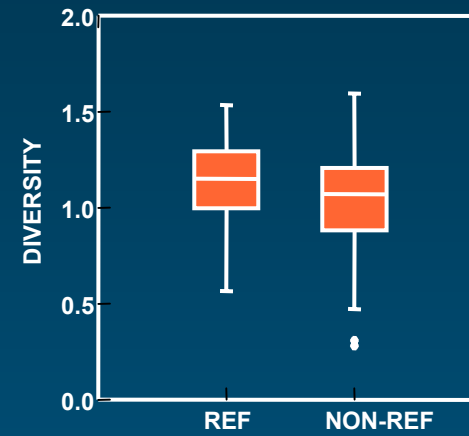
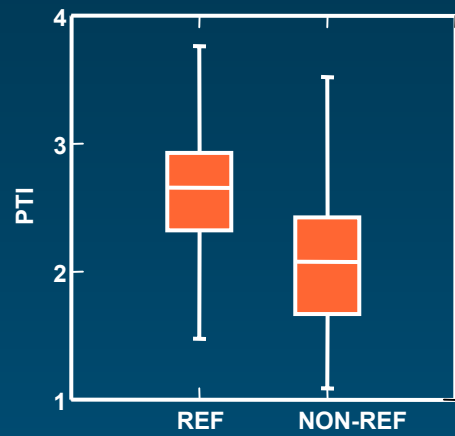
N = total number of individuals

Percent Sensitive Species

The percent sensitive species metric is the sum of the relative abundances of all intolerant (tolerance value = 4) species.

%SS = the sum of the relative abundances of all taxa with a tolerance value = 4

Original DBI Metrics



Pearson's Correlation Coefficient of Original DBI Metrics ($p < 0.01$)

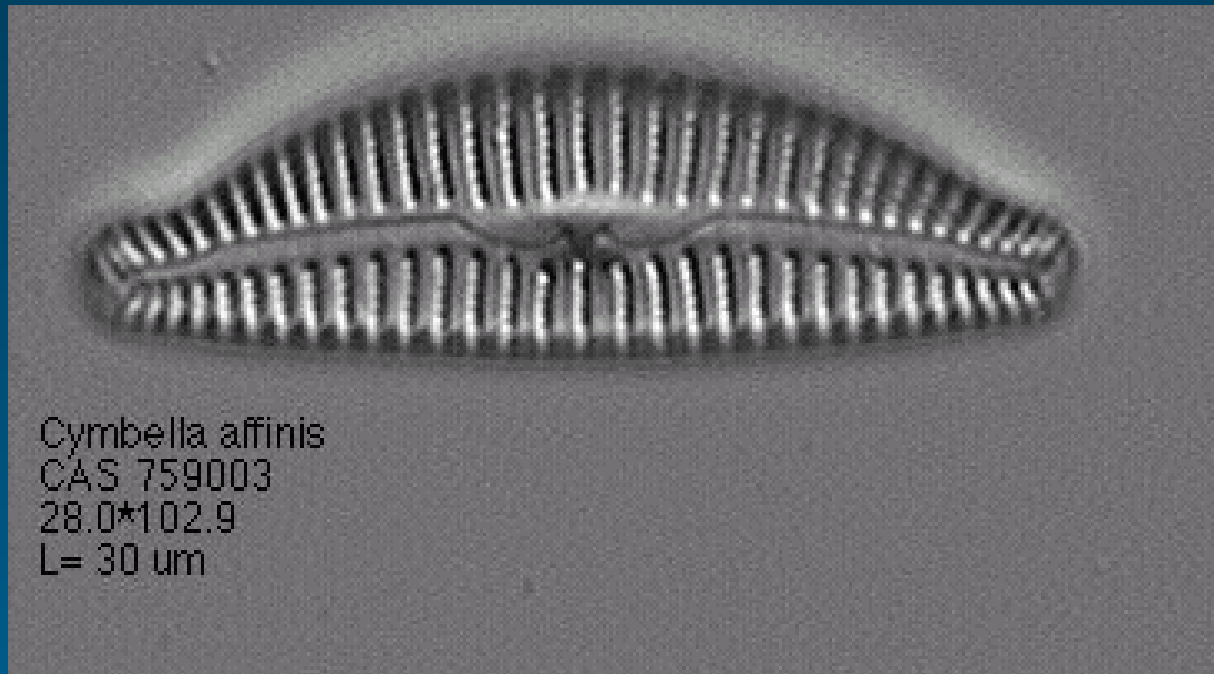
	TDTR	PTI	Diversity	%SS
TDTR	1			
PTI	0.3	1		
Diversity	0.61	0.02	1	
%SS	0.27	0.95	-0.06	1

Cymbella Group Richness (CGR)

- ◆ The total number of taxa from the following genera: *Cymbella*, *Cymbopleura*, *Encyonema*, *Encyonemopsis*, *Navicella*, *Pseudoencyonema*, and *Reimeria*.

$$\text{CGR} = \textit{Cymbella} + \textit{Cymbopleura} + \textit{Encyonema} + \textit{Encyonemopsis} + \textit{Navicella} + \textit{Pseudoencyonema} + \textit{Reimeria}$$

An Example of the *Cymbella* Group

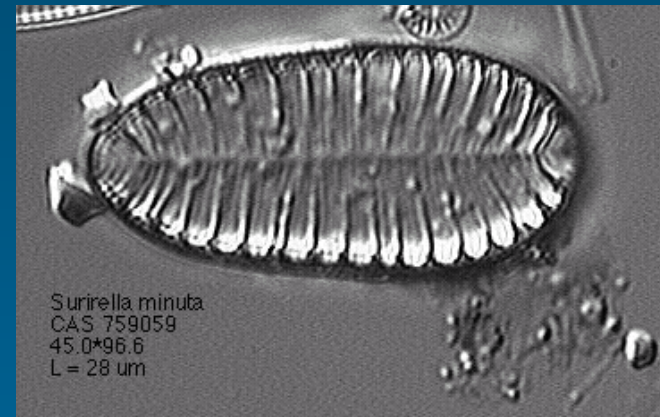
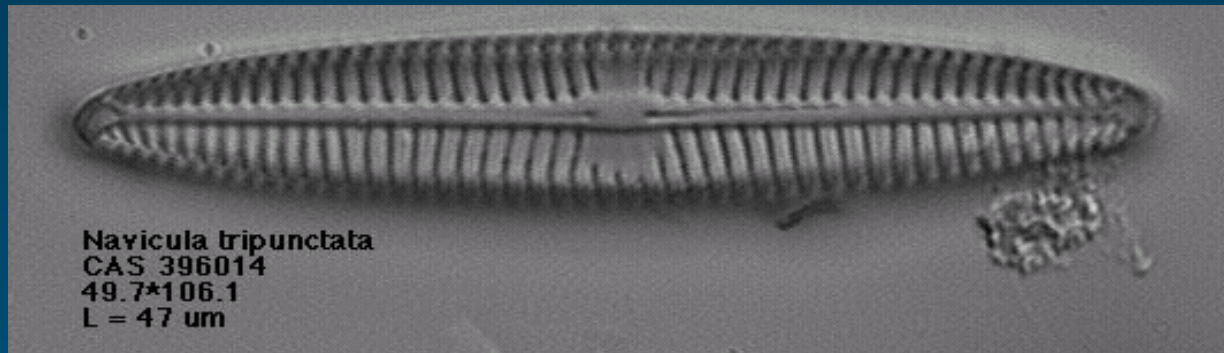


Cymbella affinis
CAS 759003
28.0*102.9
L= 30 um

% Navicula + Nitzschia + Surirella (%NNS)

%NNS = the sum of the relative abundances
of all *Navicula*, *Nitzschia*, and *Surirella*
taxa

Examples of the Genera: *Navicula*, *Nitzschia*, and *Surirella*

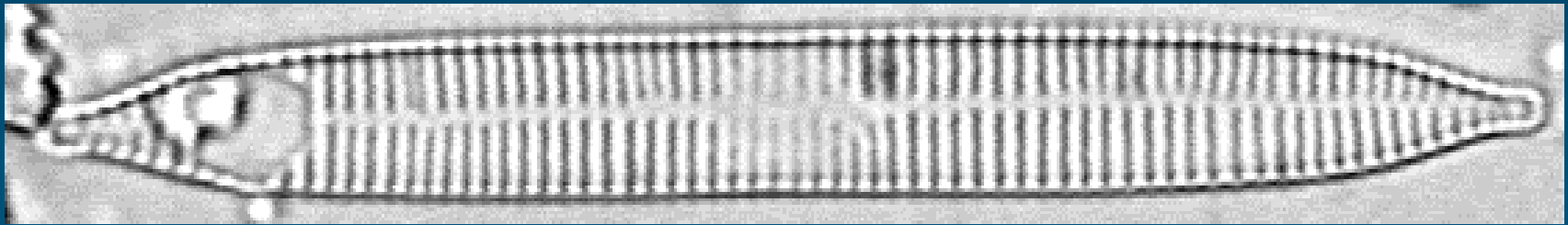


Fragilaria Group Richness (FGR)

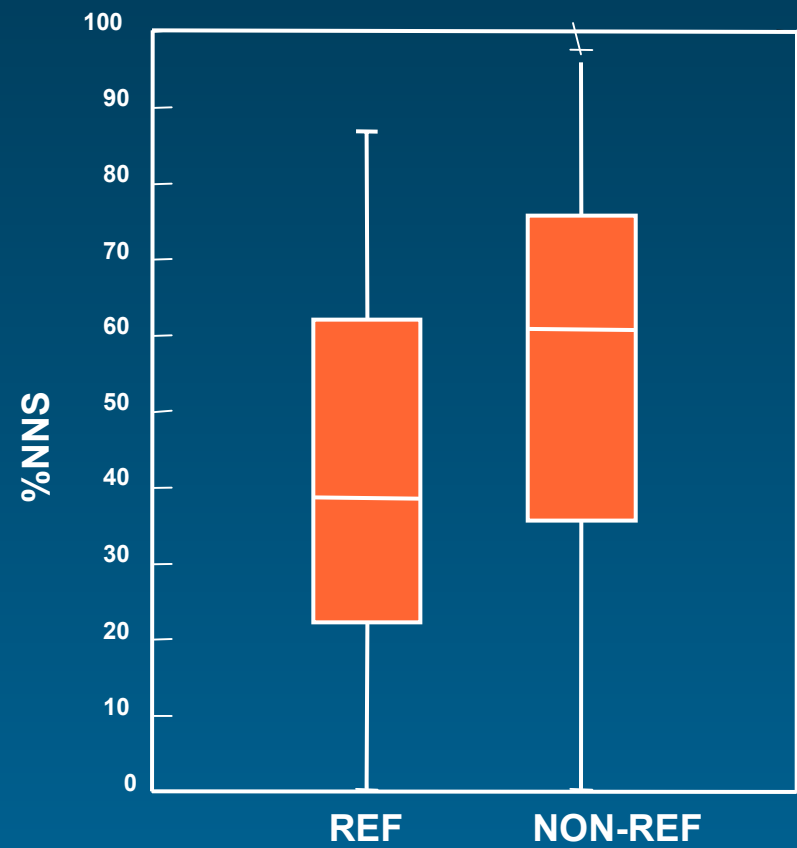
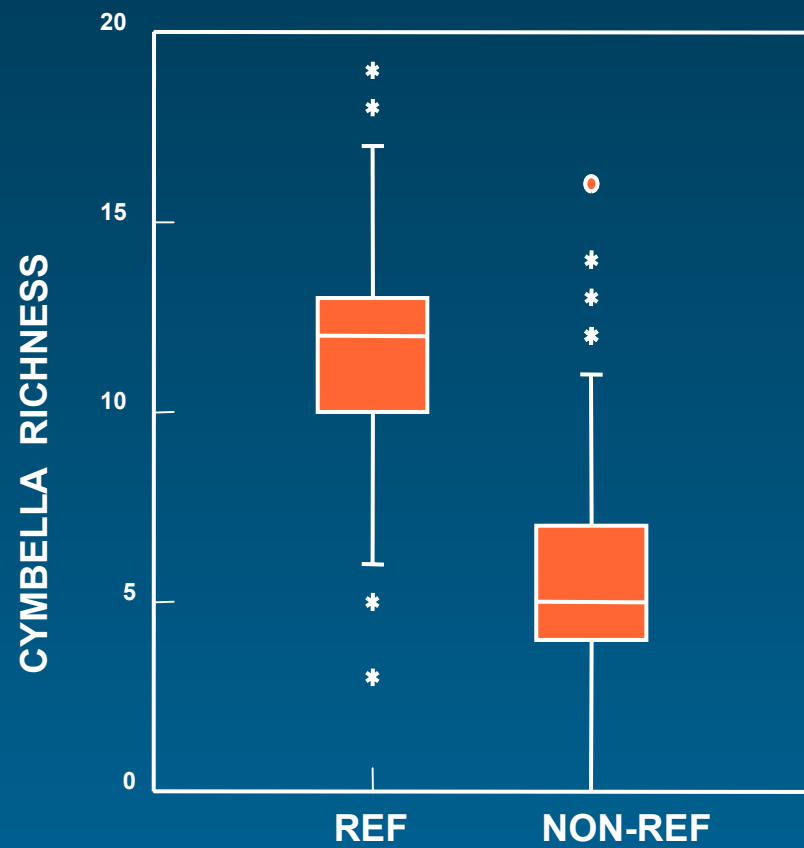
- ◆ The total number of taxa from the following genera: *Ctenophora*, *Fragilaria*, *Fragilariforma*, *Pseudostaurosira*, *Punctastriata*, *Stauroforma*, *Staurosira*, *Staurosirella*, *Synedra*, and *Tabularia*.

$$\text{FGR} = \textit{Fragilaria} + \textit{Fragilariforma} + \textit{Pseudostaurosira} + \textit{Punctastriata} + \textit{Stauroforma} + \textit{Staurosira} + \textit{Staurosirella} + \textit{Synedra} + \textit{Tabularia}$$

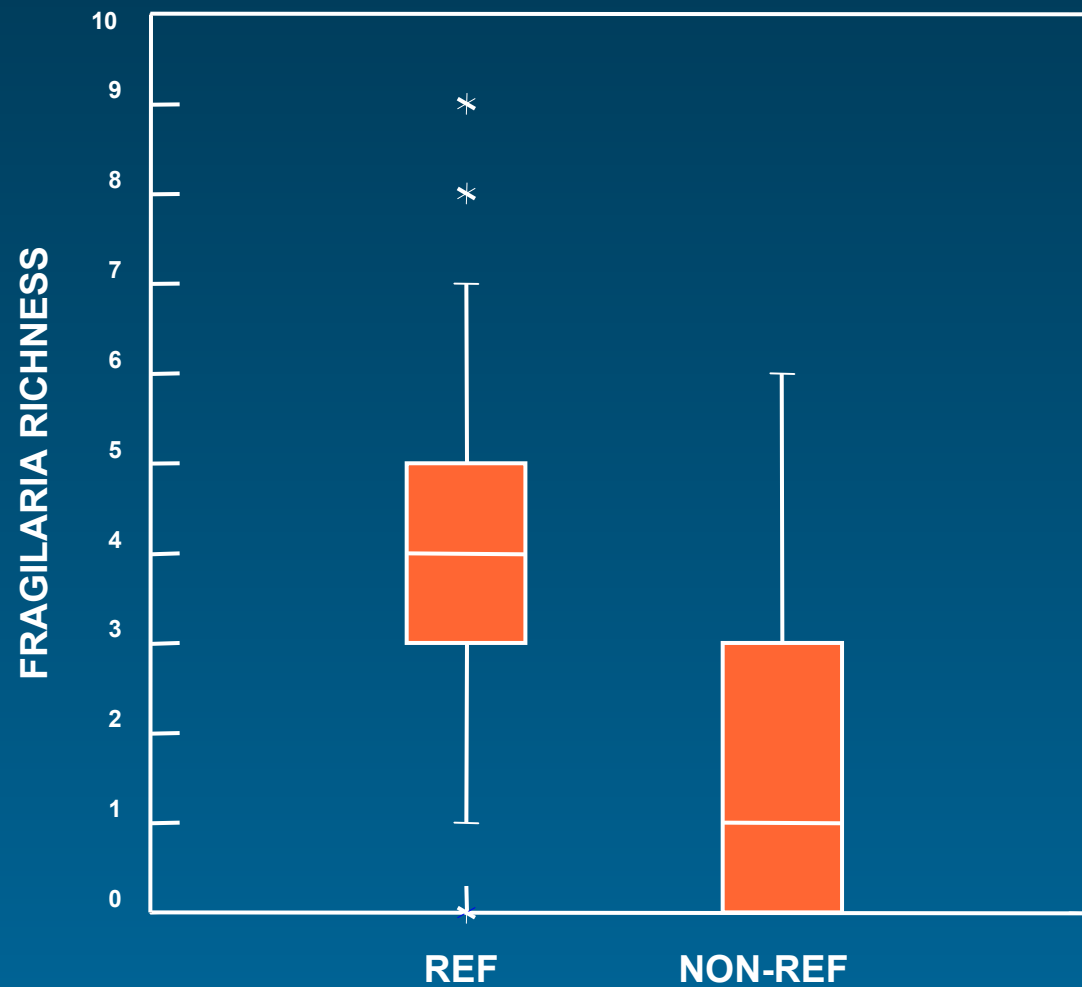
Example of the *Fragilaria* Group



New DBI Metrics



New DBI Metrics



Pearson's Correlation Coefficient for the New DBI Metrics ($p < 0.01$)

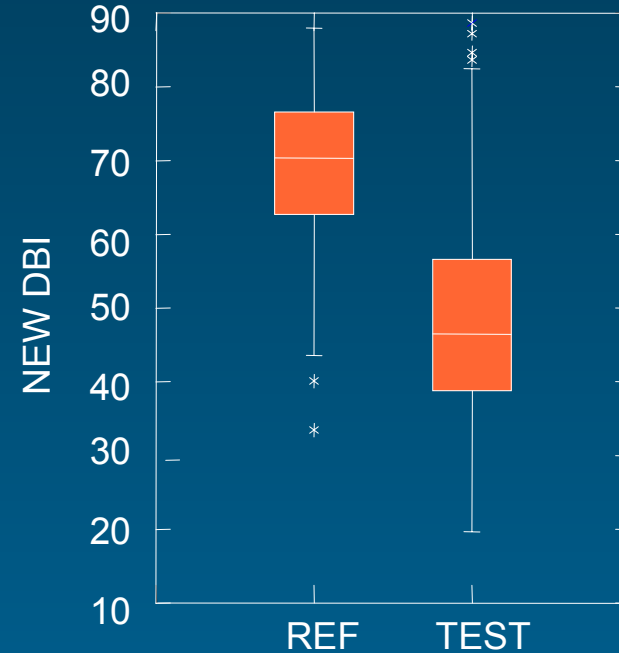
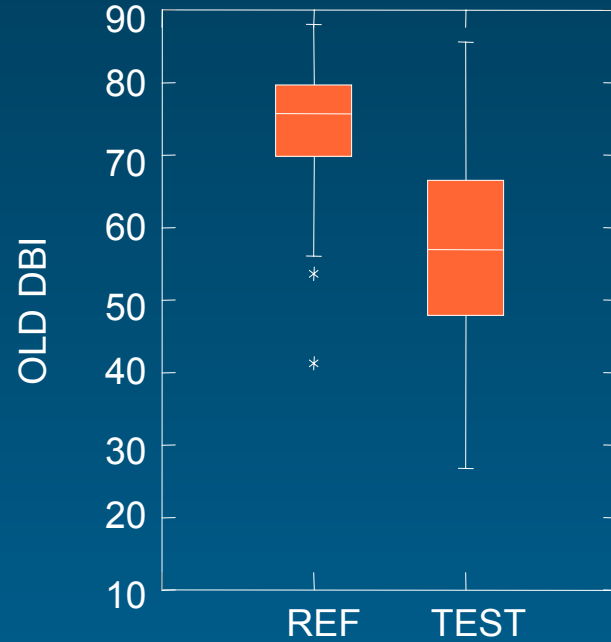
	TDTR	PTI	Diversity	Cymb Rich	% NNS	Frag Rich
TDTR	1					
PTI	0.3	1				
Diversity	0.61	0.02	1			
Cymb Rich	0.69	0.62	0.27	1		
% NNS	-0.03	-0.79	0.25	-0.42	1	
Frag Rich	0.51	0.44	0.17	0.54	-0.35	1

New DBI Metrics

The 6 DBI metrics are as follows:

- ◆ Total Number of Diatom Taxa (TNDT)
- ◆ Shannon Diversity (H')
- ◆ Pollution Tolerance Index (PTI)
- ◆ *Cymbella* Group Richness (CGR)
- ◆ *Fragilaria* Group Richness (FGR)
- ◆ % *Navicula*, *Nitzschia*, *Surirella* (%NNS)

Comparison of Old and New DBI



DBI Scoring

- ◆ For scoring the DBI, each metric is standardized to the 95th percentile of the reference distribution.
- ◆ Each metric score is based upon a 100 point scale.
- ◆ Divide the metric score by the 95th % *100.
- ◆ The total DBI score is the mean of the 6 metric scores.
- ◆ If a metric score falls above the 95th percentile, then a score of 100 is given.
- ◆ DBI scoring criteria are developed for each ecoregion, subcoregion, bioregion, etc. based upon the reference distribution.

DBI Example – Big Caney Creek

- ◆ $TNDT - (103) - 103/102 * 100 = 100$
- ◆ $Diversity - (1.41) - 1.41/1.43 * 100 = 98.60$
- ◆ $PTI - (2.863) - 2.863/3.46 * 100 = 82.746$
- ◆ $CGR - (12) - 12/13 * 100 = 92.308$
- ◆ $FGR - (6) - 6/8 * 100 = 75$
- ◆ $\%NNS - (29.91) - (100-29.91)/(100-2.6) * 100 = 71.964$
- ◆ **DBI SCORE =**
 $(100+98.6+82.746+92.308+75+71.964)/6 = 86.8$

Using the DBI for Water Quality Assessment

- ◆ DBI scores and criteria are used, in conjunction with macroinvertebrates and fish, for 305(b) site assessment purposes.
- ◆ DBI can be used to determine 303(d) successes or failures.
- ◆ Diatom taxa can indicate stressors involved in lowering the biological integrity of a site (e.g. nutrients, sediment).

Integrating Three Assemblages in Bioassessment

- ◆ All three assemblages (diatom, macroinvertebrate, and fish) are weighed equally when conducting a bioassessment of a site.
- ◆ For each assemblage, a narrative rating (Excellent – Very Poor) is derived from each index score.
- ◆ A numeric score is then assigned based upon the narrative rating (5 = Excellent – 1 = Very Poor).
- ◆ An average score is calculated to obtain an overall numeric rating for the site.

Integrating Three Assemblages in Bioassessment (Cont'd)

- ◆ The overall rating is compared to the following aquatic life use-support criteria:
 - 1. $> \text{ or } = 4.3$ Excellent Full-Support
 - 2. $4.2 - 3.6$ Good Full-Support
 - 3. $3.5 - 2.6$ Fair Partial Support
 - 4. $2.5 - 1.6$ Poor Non-Support
 - 5. $< \text{ or } = 1.5$ Very Poor Non-Support

Integrating Three Assemblages in Bioassessment (Cont'd)

- ◆ Best Professional Judgment (BPJ) will be used to determine whether assessment results of individual assemblages represent good collection effort and adequate sampling conditions.
- ◆ BPJ will also be used when one assemblage numeric score is drastically different than the other two assemblage numeric scores.

Assessing Nutrients

- ◆ Identified sites that had nutrient concentration data (ammonia (NH_3), total Kjeldahl nitrogen (TKN), nitrate (NO_3), and total phosphorus (TP)) and corresponding algae and/or macroinvertebrate data.
- ◆ Combined TKN and NO_3 into total nitrogen (TN).
- ◆ Adopted a categorical approach developed by Ohio EPA (Miltner and Rankin 1998) to examine the nutrient concentration-community structure relationship.

Assessing Nutrients (Cont'd)

- ◆ A categorical approach will allow for graphical interpretation of the response of various metrics with regard to the interaction of TN and TP concentrations.

Assessing Nutrients (Cont'd)

- ◆ All of the nutrient data in KDOW's database, Ecological Database Application System (EDAS), that had corresponding algae and/or macroinvertebrate data were utilized to determine the 25th, 50th, 75th, and 90th percentile distributions for TP (n=594) and TN (n=673).

Percentile Distribution of TP and TN Concentrations

◆ 25 th %	0.014 mg/l TP	0.386 mg/l TN
◆ 50 th %	0.045 mg/l TP	0.860 mg/l TN
◆ 75 th %	0.163 mg/l TP	1.763 mg/l TN
◆ 90 th %	0.710 mg/l TP	4.178 mg/l TN

TP (n=594)

TN (n=673)

Nutrient Categories (Adapted after Miltner and Rankin 1998)

- ◆ Bioassessment sites were placed into one of six categories based upon the percentile rankings for TP and TN at those sites.

Nutrient Categories (Adapted after Miltner and Rankin 1998)

- ◆ Category 1 both \leq TP₂₅ TN₂₅
- ◆ Category 2 either \leq TP₅₀ TN₅₀
- ◆ Category 3 \leq TP₇₅, $<$ TN₉₀
- ◆ Category 4 $>$ TP₇₅, $<$ TN₉₀
- ◆ Category 5 both $>$ or $=$ TP₉₀ TN₉₀
- ◆ Category 6 NH₃ $>$ or $=$ 1.0 mg/l (toxic)

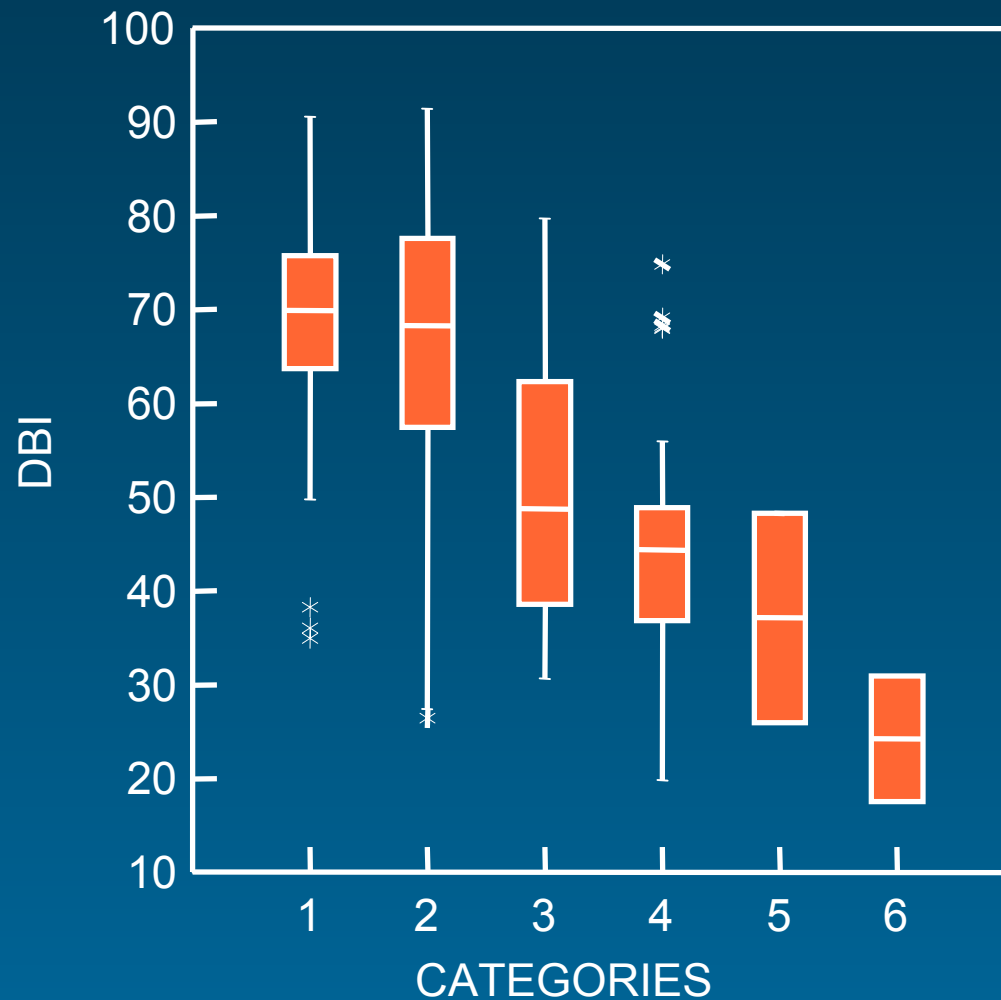
Using Algal Data to Assess Nutrients

- ◆ A data set of paired diatom and nutrient samples (n=254) was evaluated where DBI and component metrics were graphed and correlated with log transformed nutrient concentrations.

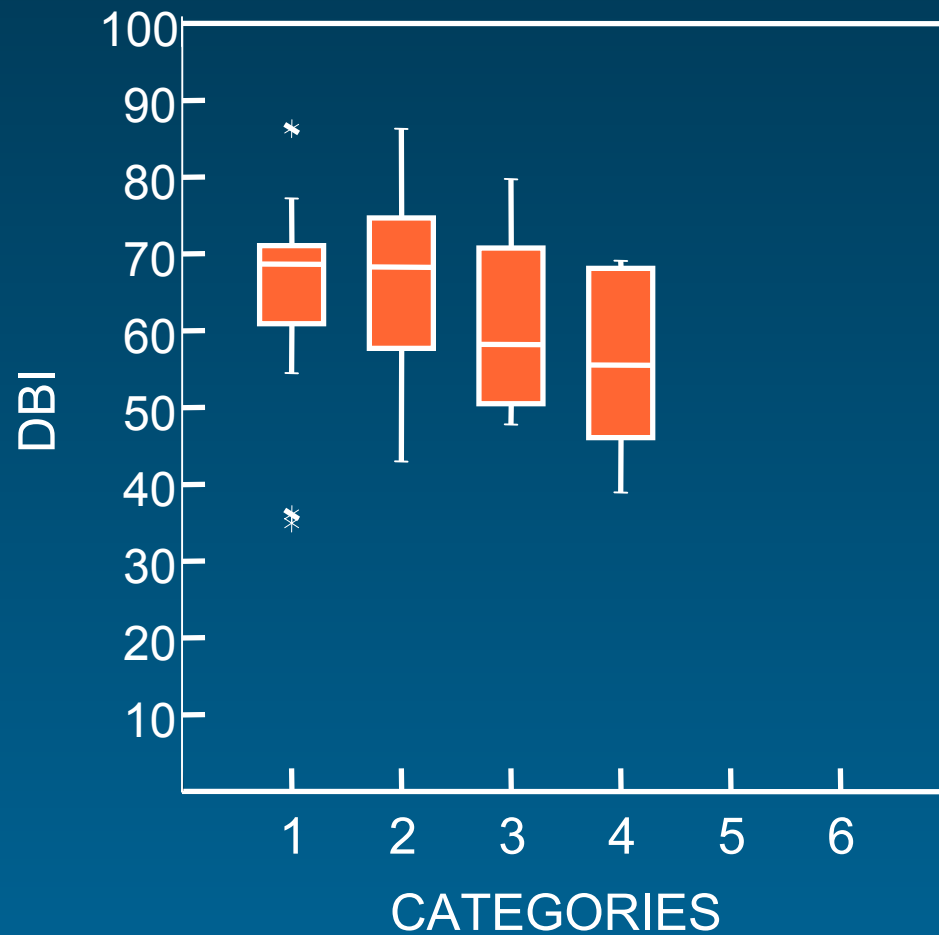
Pearson Correlation Matrix of Nutrients and Diatom Metrics ($p < 0.01$)

	TP	TN	TN*TP
TNDT	-0.21	-0.30	-0.30
Diversity	0.17	0.08	0.13
PTI	-0.46	-0.57	-0.60
CGR	-0.42	-0.44	-0.49
FGR	-0.42	-0.44	-0.50
%NNS	0.51	0.54	0.61
%NUTTOL	0.43	0.52	0.55
DBI	-0.48	-0.54	-0.59

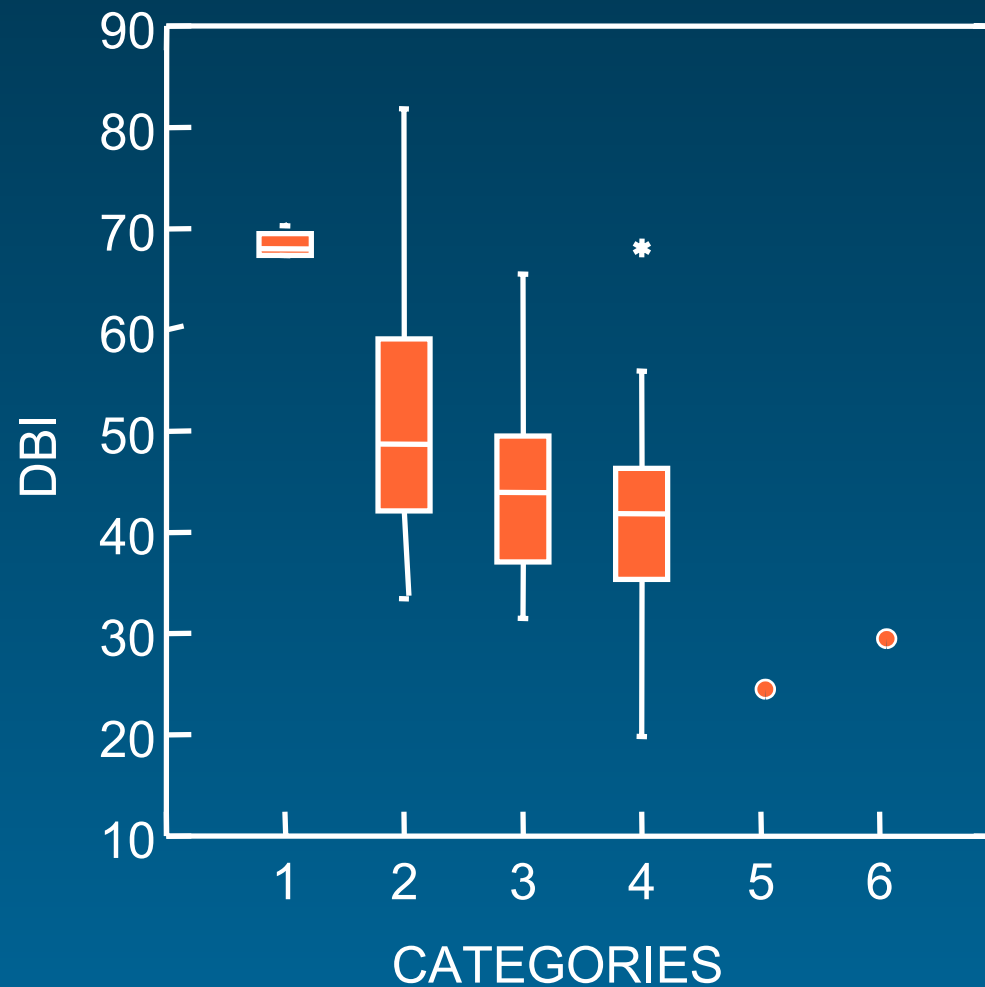
Statewide DBI vs. Nutrient Categories



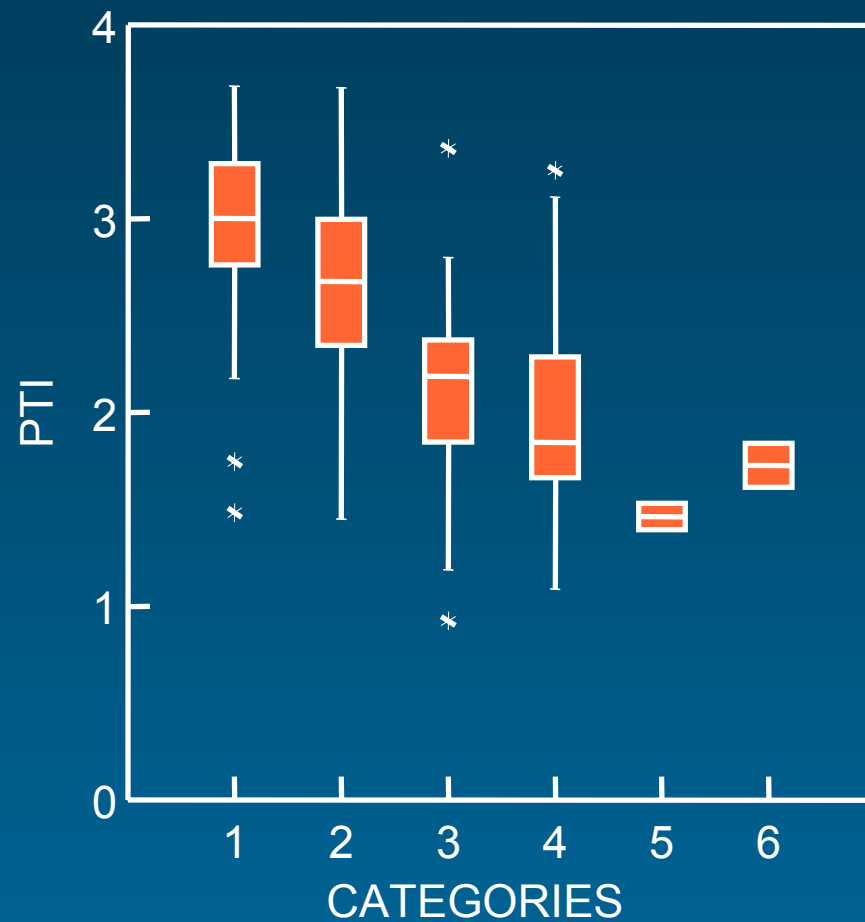
Pennyroyal DBI vs. Nutrient Categories



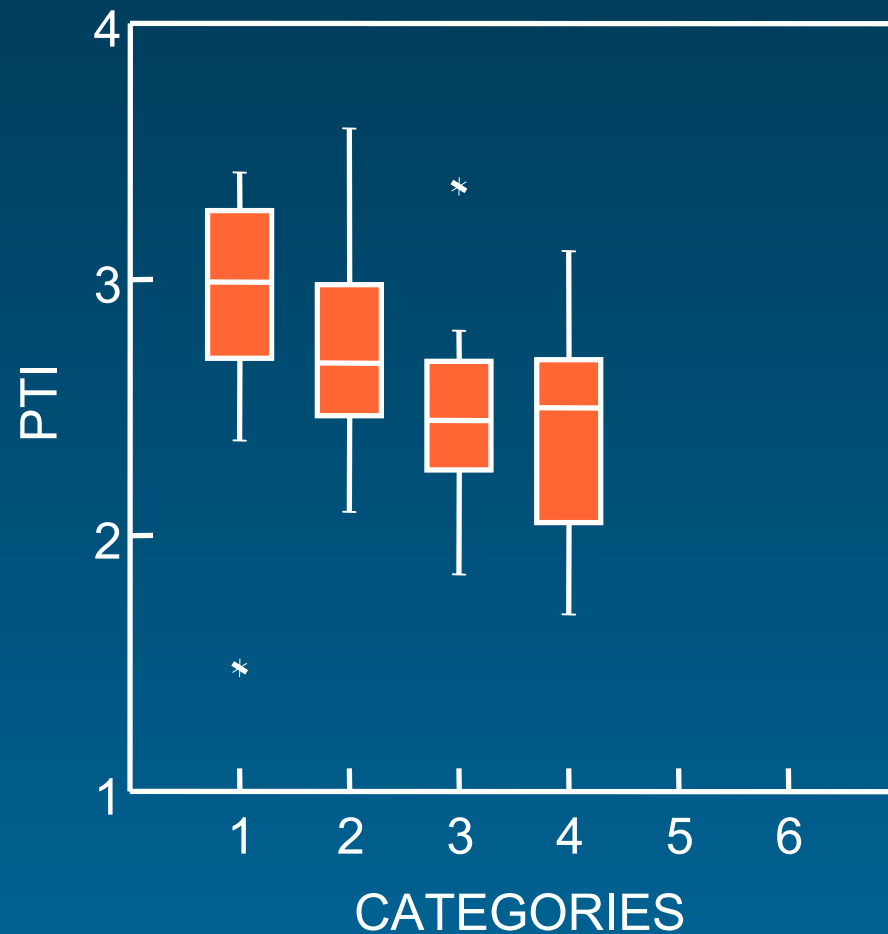
Bluegrass DBI vs. Nutrient Categories



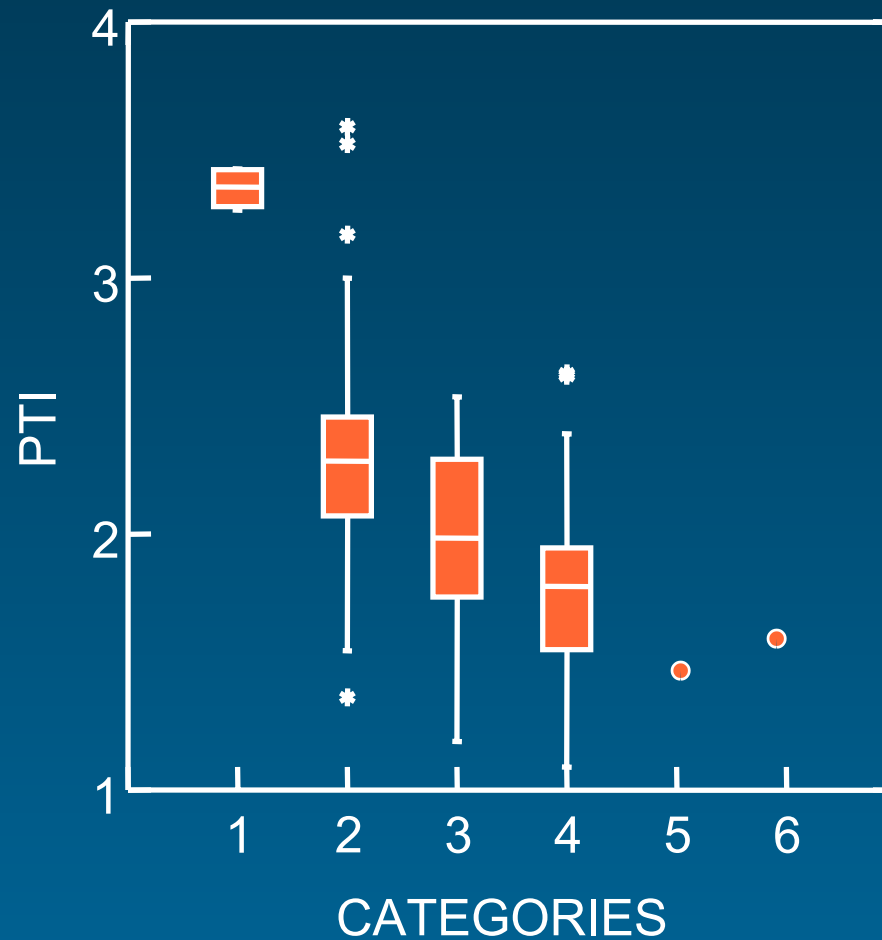
Statewide PTI vs. Nutrient Categories



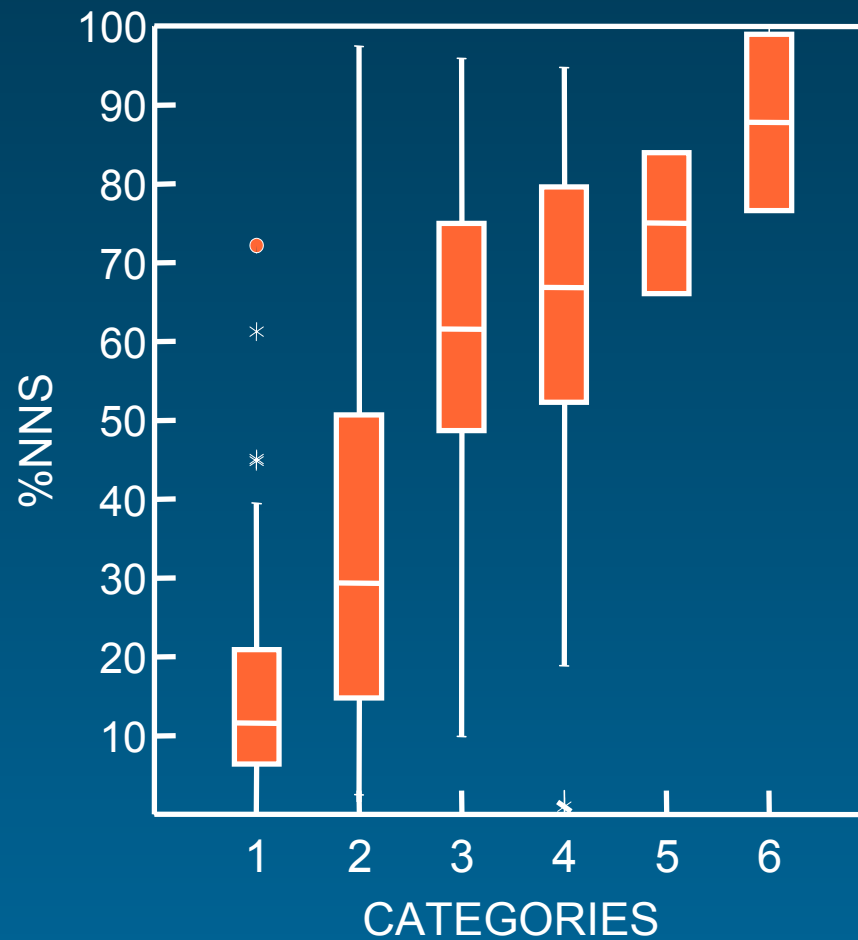
Pennyroyal PTI vs. Nutrient Categories



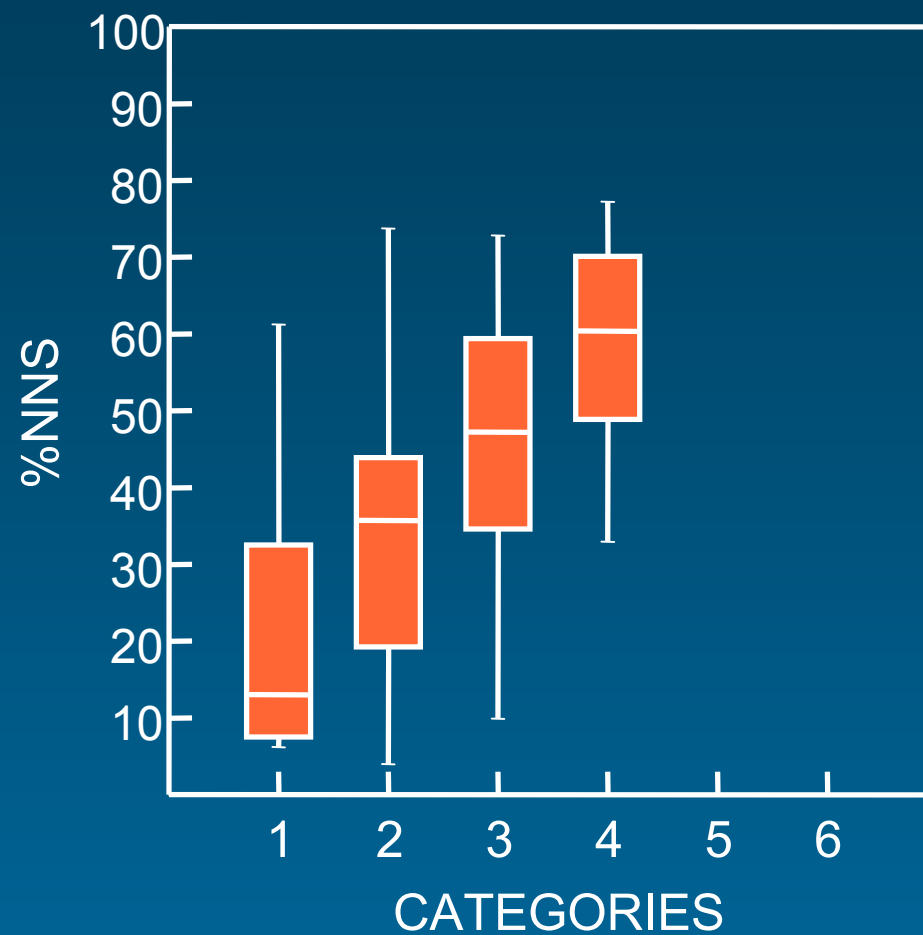
Bluegrass PTI vs. Nutrient Categories



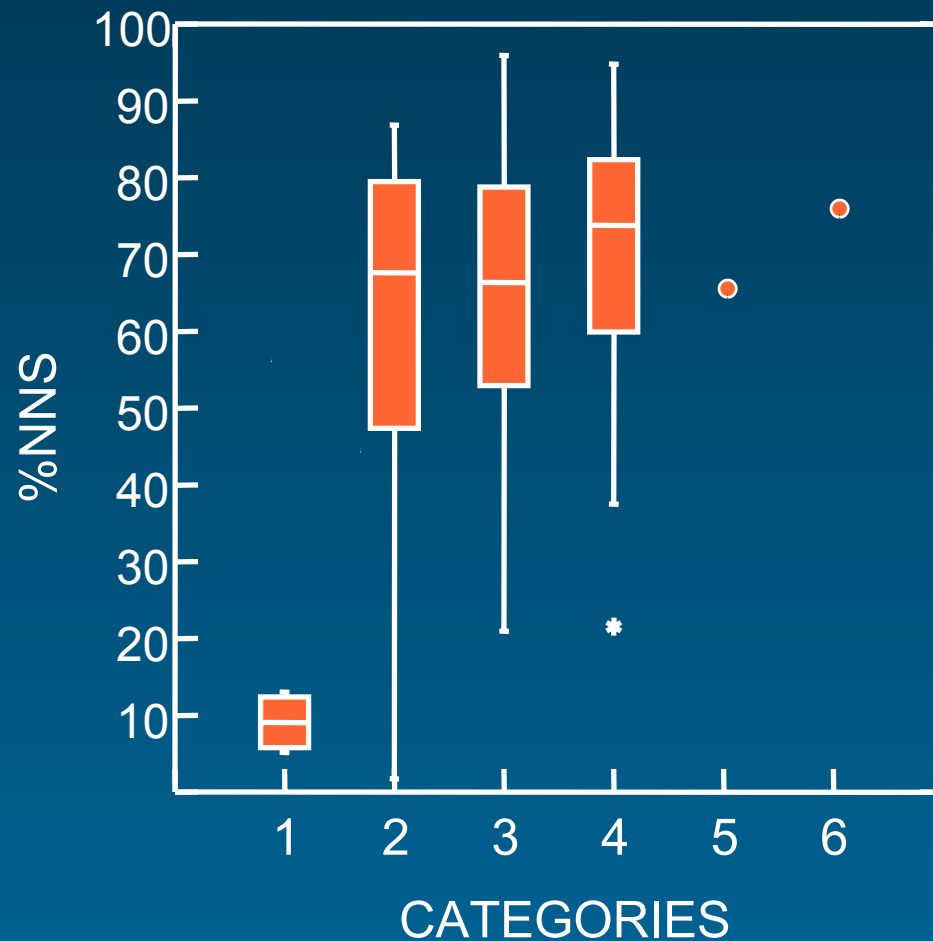
Statewide % NNS vs. Nutrient Categories



Pennyroyal %NNS vs. Nutrient Categories



Bluegrass %NNS vs. Nutrient Categories



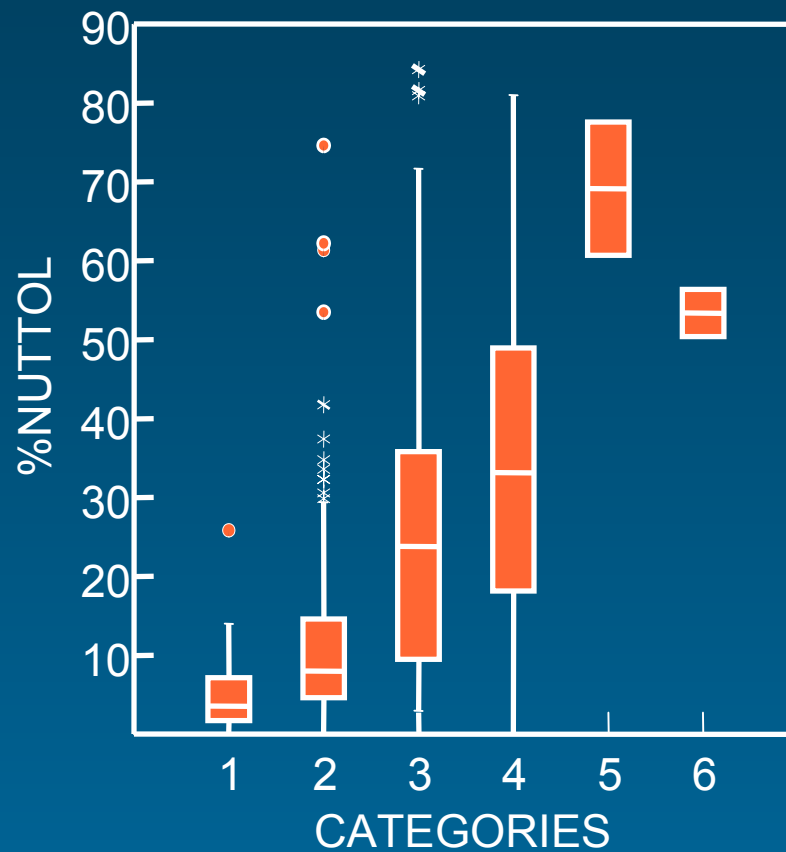
%NUTTOL vs. Nutrient Categories

- ◆ % Nutrient Tolerant Taxa (%NUTTOL) is another assessment tool that can be used to identify nutrient enrichment as a pollution source.
- ◆ This metric combines the relative abundances of the top ten nutrient tolerant diatom taxa.
- ◆ Each of these taxa have a PTI tolerance value of one (most tolerant) and are among the most commonly encountered taxa in nutrient-rich waters.

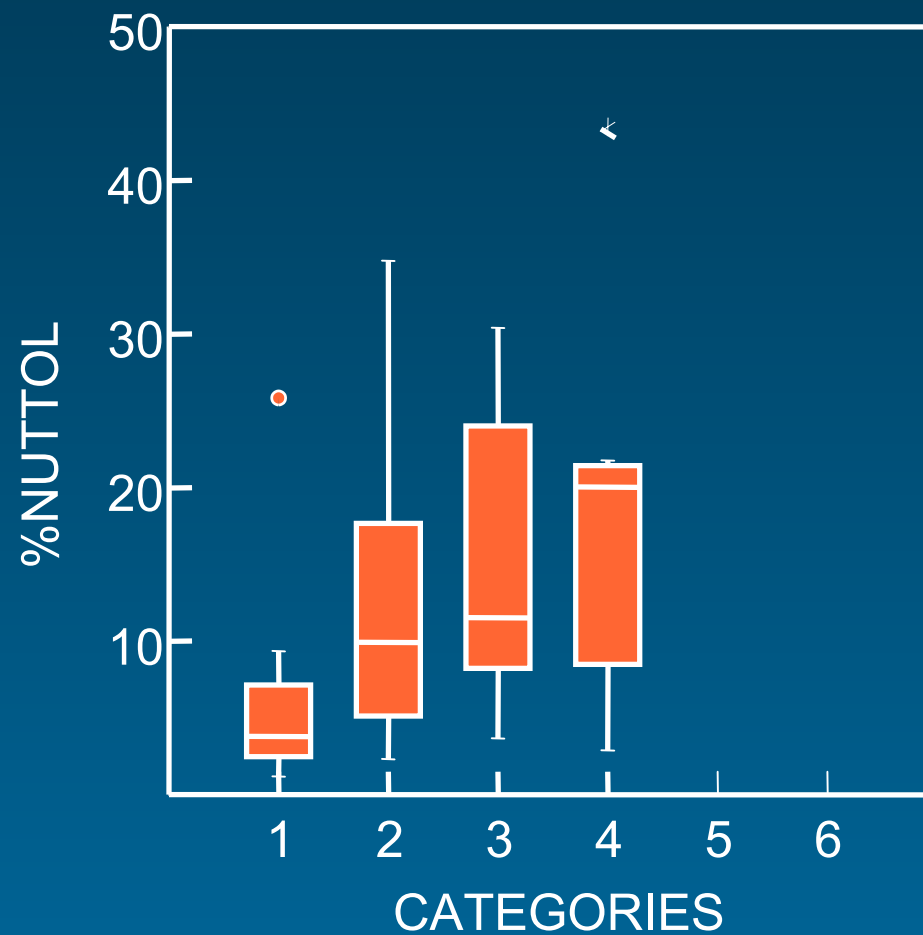
%NUTTOL vs. Nutrient Categories (Cont'd)

%NUTTOL=the sum of the relative abundances of *Gomphonema parvulum*, *Navicula cryptocephala* var. *veneta*, *Navicula minima*, *Navicula seminulum*, *Navicula subminuscula*, *Nitzschia amphibia*, *Nitzschia filiformis*, *Nitzschia frustulum*, *Nitzschia palea*, and *Stephanocyclus meneghiniana*

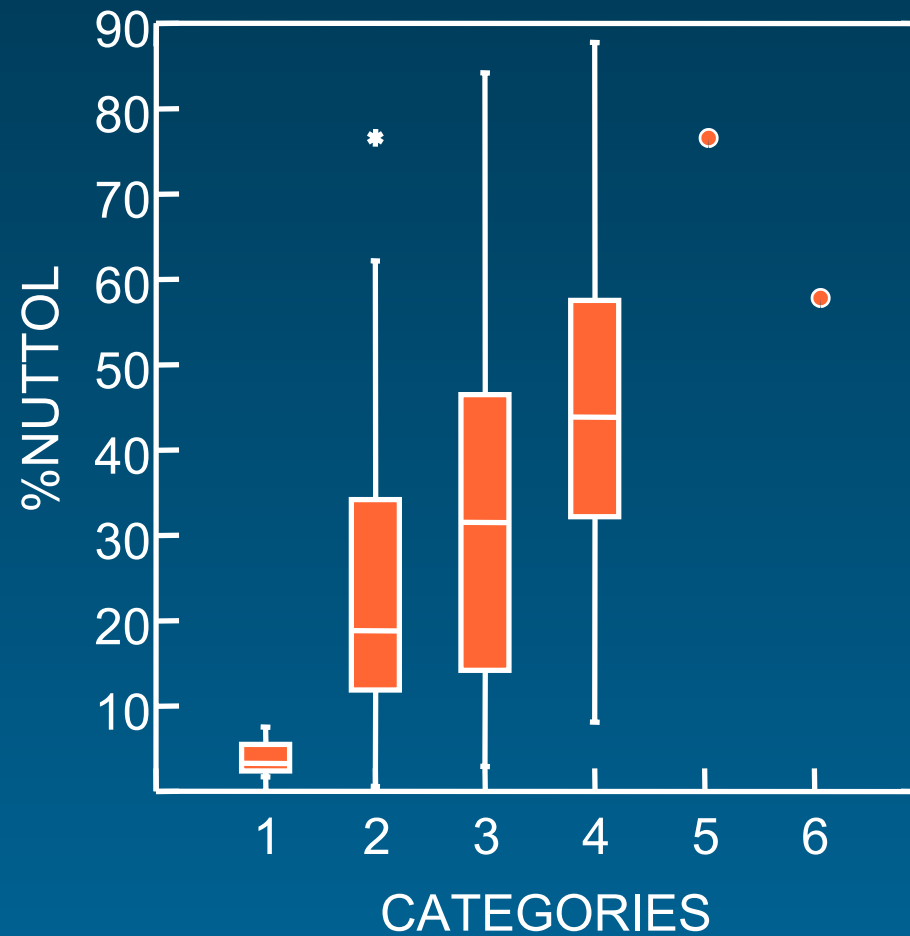
Statewide %NUTTOL vs. Nutrient Categories



Pennyroyal %NUTTOL vs. Nutrient Categories



Bluegrass %NUTTOL vs. Nutrient Categories



Future Considerations

- ◆ Need more nutrient concentration data collected in association with diatom and macroinvertebrate data, especially in some bioregions where associated data are lacking.
- ◆ Large river methodology and index development (USEPA lab in Cincinnati).
- ◆ Use the categorical approach to analyze the influence of other stressors (ex. Conductivity) on the diatom community.

Future Considerations (Cont'd)

- ◆ Testing the viability of using non-diatom metrics to enhance DBI.
- ◆ Use viewing bucket methodology to semi-quantitatively assess algal division representation and coverage in the field.
- ◆ Determine whether nutrient concentration thresholds exist where algal community structure is compromised.

References

Miltler, R.J. and E.T. Rankin. 1998. Primary nutrients and the biotic integrity of rivers and streams. *Freshwater Biology* 40: 145-158.

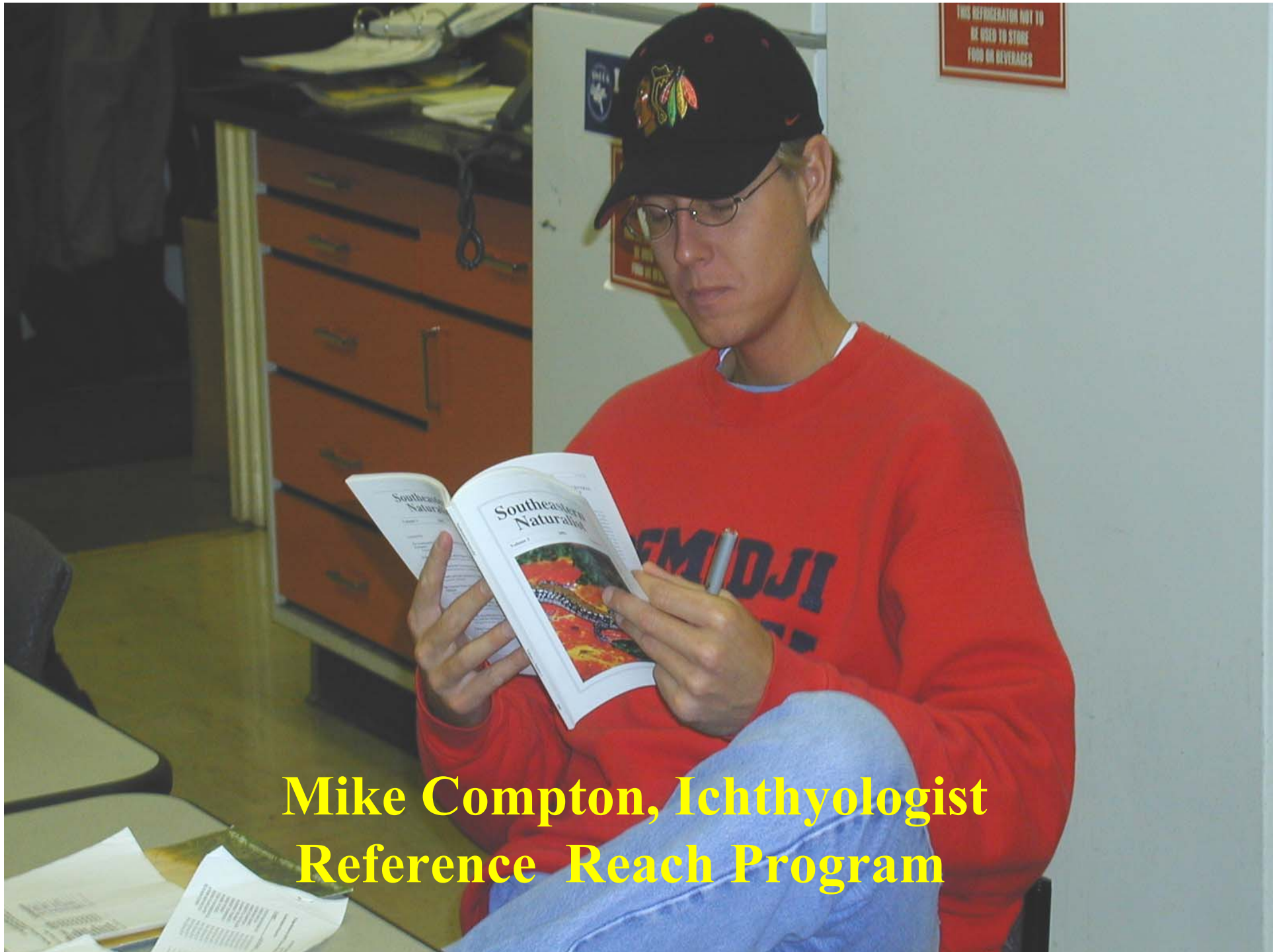
Resource Materials

Standard Operating Procedures Manual:

– www.water.ky.gov/sw/swmonitor/sop

Diatoms and Nutrients Manual (Draft):

- john.brumley@ky.gov



Mike Compton, Ichthyologist
Reference Reach Program



Greg Pond, Macroinvertebrate Specialist
Reference Reach Program

Special Thanks

Paulette Akers



Special Thanks

- ◆ Lythia Metzmeier – friend and DOW colleague; originally developed DBI in early 90's
- ◆ Stephan Porter – original phycologist at DOW
- ◆ Dr. Jan Stevenson – support and guidance, especially when he was at U of L